

Mandle

THE COLLEGE

Laboratory of Microbiology Botanical Laboratory 38th St. and Woodland Ave. June 17, 1949

Dr. Joshua Lederberg
Department of Genetics
University of Wisconsin
College of Agriculture
Madison 6, Wisconsin

Dear Dr. Lederberg:

I was very stimulated this morning by the arrival of your letter and the four cultures of A. tumefaciens. Many thanks for your generosity. The cultures came through in good condition.

Mr. Gordon's manuscript was very informative and I wish you would thank him for me.

In reference to your letter, I am glad to know of the Aniline Blue medium. Riker has proposed other media containing dyes about which Dr. Braun told me, but they were not too reliable. I will try your suggestion and use the Aniline Blue.

The mutants you sent, (GI and GII) are similar to the only stable one I have, in that the deficiency is of a Sulphur compound or ion. My stable one is a methionine less strain. It showed no growth in any of the amino acids tested including cystein, L (-) cystine, and glutathione. However, sulfide ions alone were never tested but I strongly suspect that these too would prove negative. It might be possible to use one of your strains together with my M7 (Methionine less) in stars and make isolations from them. You made no mention of star formation with the A6 strain so I presume that you had no difficulty with it. Dr. Braum and myself have experienced some difficulty in obtaining good stars with the A6 so for that reason we chose the B2, the IIBV7, and the T37 strains for our work. The M7 is of IIBV7 origin. This is a nice strain to work with because of its ease in forming stars and its relative stability; it throws off fewer rough colonies than the other strains with which we have worked. There is another feature to this strain in as much as we have a non virulent (for almost all of the hosts that the sister cell isolate shows virulence) strain which is an apparent stable mutation. This could be used as an additional character.

I am in the process of attempting to isolate a mutant of the  $B_2$  strain. It will be several weeks yet before I will have any indication of the outcome of the present search.

The work with the tetrazolium reaction is more than ever confusing. When I talked to you it was thought that the "cofactor" would be in the peptone and could be tested for with cell suspensions. Latter work has served only to further confound me. It seems that in addition to pH and peptone, casein hydrolysate, and yeast extract will also aid the reduction. When the organisms were mixed with various substrates in Thunberg tubes all substrates and the minimal medium allowed reduction to the red formazan. This served to further complicate the findings. It is apparently not as simple as was first thought.

If you and/or Mr. Gordon are planning to continue the work with tumefaciens please let me know so that I may send you strains and such information as I might have.

I appreciate the interest and generosity you have shown. Please give my regards to your wife and thank you again for your help.

Sincerely yours,

Robert J. Mandle

R.J. mandle

RJM/aep